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Dorsal striatum mediation of cocaine-seeking after withdrawal from short or long daily access cocaine self-administration in rats

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Abstract

Accumulating evidence has suggested that prolonged use of cocaine may lead to progressive neuroadaptations proceeding from ventral to more dorsal areas of the corpus striatum. We have previously found that reversible inactivation of the dorsolateral caudate/putamen (dlCPu) significantly attenuated cocaine-seeking in rats following chronic cocaine self-administration and withdrawal. Since the cumulative amount of cocaine intake and the time course of withdrawal emergent patterns have been previously shown to alter subsequent cocaine-seeking, the current study investigated the role of the dlCPu in cocaine-seeking after differing access periods of cocaine self-administration and abstinence time points. Rats were catheterized and implanted with infusion cannulae in the dlCPu, trained on cocaine self-administration (0.2 mg/50 μ l/infusion), and then allowed to self-administer cocaine for 1 or 6 h daily sessions. After the final session, animals underwent three separate tests of cocaine-seeking in the self-administration context at days 1, 14, and 60 of abstinence immediately following bilateral infusion of baclofen-muscimol or vehicle into the dlCPu. While inactivation of the dlCPu by baclofen-muscimol resulted in reduced cocaine-seeking in both groups, the degree of inhibited responding varied with access history and withdrawal time point. While these data support a role for dorsal striatal regions in cocaine-seeking, greater previous cocaine intake did not lead to a greater dependence on intact dlCPu function for cocaine-seeking after abstinence.

Keywords

caudate-putamen; cocaine; relapse; self-administration; striatal

1. Introduction

Studies on the mesostriatal regulation of drug reward have primarily focused on the ventral tegmental area to nucleus accumbens pathway of the brain, particularly the ascending

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mesolimbic dopamine (DA) projections [1,2,3]. While the evidence for a primary role of the mesolimbic pathway in mediating drug reward is fairly unequivocal, recent data suggests that prolonged intake of abused drugs, particularly cocaine, results in progressive changes across striatal regions. Specifically, it has been proposed that neuroadaptations within the dorsal striatum (i.e., caudate-putamen) come to exert greater control over drug-seeking [4,5,6]. As drug use progresses over time, ventral striatal areas may maintain less control over drug-seeking behavior [7], reflected by a relative shift from action–outcome to habitual stimulus-response mediated behaviors [8].

The idea of a ventral to dorsal striatal shift has come from several convergent lines of research. In rat self-administration models of cocaine addiction, increased extracellular DA in the dorsal striatum has been reported during responding for a cocaine-associated cue [9], while DA receptor antagonist infusion blocked responding for a cocaine-associated cue under a second-order schedule of reinforcement when placed into the dlCPu [10], or via a functional disconnection of the nucleus accumbens core and dlCPu [11]. Finally, in an abstinence relapse model in rats with a history of cocaine self-administration, inactivation of the dlCPu or the substantia nigra pars compacta attenuated context-induced cocaine-seeking [12,13].

In monkeys that self-administered cocaine, subjects with increased cocaine access and intake showed more pronounced patterns of dorsal striatum changes as seen by increased DA transporter levels [14] and decreased functional activity as measured by deoxyglucose binding [7]. In brain imaging studies in humans, cocaine-dependent subjects showed increased DA release in the dorsal (caudate and putamen), but not ventral striatum as determined by [¹¹C]raclopride binding [15,16]. Furthermore, the magnitude of dorsal striatal DA binding correlated with self-reported craving. Such changes point to a critical role of the caudate-putamen in craving for cocaine and as a site for long term neuroplasticity after repeated cocaine use.

In animal models of relapse, re-exposure to a context previously associated with drug self-administration results in robust drug-seeking, as evidenced by responding on a previously drug-reinforced operandum [12,17]. The history of drug intake is a critical factor in the motivation to seek cocaine at a later time point, since extended daily access to cocaine during self-administration enhances subsequent cocaine-primed [18,19,20] and conditioned-cue induced reinstatement [18] when compared to rats with shorter cocaine access histories. Another important factor in subsequent drug-seeking seems to be the duration of time after chronic self-administration. Reinstatement of cocaine-seeking produced by discrete drug-paired cues has been shown to increase over time after cessation of cocaine self-administration [21], an effect known as “incubation of craving” [22]. This enhanced responding has been seen after varied periods of withdrawal and subsequent extinction trials [22], and during context-induced responding after varied periods of abstinence [23]. However, studies of reinstated or renewed drug-seeking have generally not examined brain regions that may regulate drug-seeking after different cocaine daily access conditions.

Here, we investigated the contribution of the dlCPu to cocaine-seeking after different lengths of daily cocaine self-administration and varied periods of abstinence. We predicted that reversible inactivation of the dlCPu would attenuate context-induced cocaine-seeking to a greater degree in animals with a history of long daily access when compared to animals with short daily access.

2. Materials and methods

2.1. Subjects

Male Sprague–Dawley rats (Charles River Laboratories, Wilmington, MA) weighing 275–300 g were individually housed on a 12 h reverse light cycle, with lights on at 19:00. Housing and care of the rats were carried out in accordance with the National Institute of Health Guide for the Care and Use of Laboratory Animals (NIH Publications No. 80-23). Formal approval to conduct the experiments was obtained from the MUSC IACUC. Steps were taken to minimize animal pain and suffering and limit the total number of animals used in these studies.

2.2. Surgery

Anesthesia consisted of IP injections of ketamine (66 mg/kg), xylazine (1.3 mg/kg), and equithesin (0.5 ml/kg). Ketorolac (2.0 mg/kg, IP) was given just prior to surgery as an analgesic. One end of a silastic catheter was inserted into the external right jugular and secured with 4.0 silk sutures. The other end ran subcutaneously and exited from a small incision just below the scapula to provide access to an external port for IV drug delivery. Immediately after catheter surgery, animals were placed in a stereotactic frame (Stoelting, Wood Dale, IL) and implanted with stainless steel guide cannulae (26 gauge, Plastics One, Roanoke, VA) just dorsal to the dlCPu (coordinates in mm: +1.2 anterior, ± 3.8 medial-lateral, -3.4 ventral) based on a rat brain atlas [24]. Cannulae were secured to the skull using jeweler's screws and dental acrylic and stylets were placed into the cannulae to prevent blockage. An antibiotic solution of cefazolin (10 mg/0.1 ml) was given post surgery and during recovery along with 0.1 ml of 70 U/ml heparinized saline. During self-administration, rats received an IV infusion (0.1 ml) of 10 U/ml heparinized saline before each session. After each session, catheters were flushed with cefazolin and 0.1 ml of 70 U/ml heparinized saline. Catheter patency was periodically verified with methohexital sodium (10 mg/ml IV), a short-acting barbiturate that produces a rapid loss of muscle tone.

2.3. Cocaine self-administration

All self-administration sessions occurred during the animals' dark cycle and were conducted 6 days per week. Before each session, catheters were connected to flexible polyethylene tubing enclosed in a spring leash (Plastics One) and attached to a swivel (Instech, Plymouth Meeting, PA) mounted on top of the chamber. Cocaine hydrochloride (National Institute on Drug Abuse, Research Triangle Park, NC) was dissolved in saline, filtered, and delivered by active lever responding at a dose of 0.2 mg/50 μ l infusion throughout the experiment. Each lever response resulted in a 2 s infusion and 20 s timeout period, during which time lever responses were recorded, but no drug was administered. No discrete stimuli (e.g., tone or light) were presented with active lever responses, and responses on the inactive lever were recorded, but had no scheduled consequences. As inactive lever responding was uniformly very low across all sessions and did not reveal any differences between groups, these data are not presented. Animals were required to reach a criterion of at least 10 infusions/session, which took between 2–4 days. Upon achieving this criterion, all animals first experienced six consecutive days of 2 h/day sessions. Animals were then divided into short access (1 h/day; $n=20$) or long access (6 h/day; $n=23$) for an additional 15 days of self-administration. We chose these regimens based on previous studies in rats that have found behavioral and neurobiological differences between animals with a history of short vs. long daily access to cocaine [18,25,26].

2.4. Context-induced cocaine-seeking

Rats experienced abstinence from cocaine for a period of 60 days. On days 1, 14, and 60 of abstinence, animals received intra-dlCPu infusions of B/M or vehicle immediately before a test session in the self-administration chamber to assess cocaine-seeking (context test day). The order of B/M and vehicle infusions were counterbalanced across rats and test sessions. Each rat received only B/M or vehicle on a given test day (i.e., rats did not experience both B/M and vehicle injections on each test day). Rats remained in their home cages during abstinence, except that one week prior to test days 14 and 60, they were transported to an alternate environment room and placed in Plexiglas boxes for 2 h at the same time of day. This procedure was designed to mitigate the potential stress of not being handled for an extended period of time [12,13]. On post-cocaine days 1, 14 and 60, the rats received intra-dlCPu infusion of baclofen and muscimol (B/M) or vehicle (phosphate-buffered saline) prior to testing.

2.5. Intracranial infusions

Just prior to tests of cocaine-seeking after abstinence, bilateral infusion cannulae (33 gauge) were inserted to a depth of 2 mm below the tip of the guide cannulae, which allowed for solution to traverse into the dlCPu along the infusion track. In order to acclimate rats to the infusion procedure, injection cannulae were bilaterally inserted into the guide cannulae for 3 min (no infusions) before placement into the self-administration chamber on the last self-administration day and on post-cocaine day 59. On test days, B/M (1.0/0.1 mM) or vehicle were infused (0.6 µl/side) immediately before being placed into the previously cocaine-paired (i.e., self-administration) context. Previous studies have shown that this amount and concentration of B/M attenuates cocaine-seeking when infused into the dlCPu [12,13]. Infusions occurred via polyethylene tubing connected to gastight Hamilton syringes (10 µl; Hamilton, Reno, NV) set in an infusion pump (PHD 2000, Harvard Apparatus, Holliston, MA) and delivered over a 2 min period, and injectors were left in place for an additional minute.

2.6. Histology

At the end of behavioral testing, rats received an overdose of equithesin and were transcardially perfused with phosphate buffered saline and 10% formaldehyde. After extraction, brains were stored in 10% formaldehyde before sectioning on a vibratome. Coronal sections (75 µm) were mounted onto gelatin-coated slides, stained with cresyl violet, and cannula placement confirmed by examining slides under a light microscope [24]. Animals were excluded from data analysis if the most ventral part of the infusion cannula track fell outside the targeted structure.

2.7. Data analysis

Repeated measures analysis of variance (ANOVA) was used to analyze lever responses and cocaine intake over the period of self-administration, followed by post hoc comparisons (Tukey test) for individual days. Data were also analyzed for the first hour of responding and cocaine intake during self-administration for the 1 h and 6 h/day groups. Responses during context-induced cocaine-seeking were analyzed by three-way ANOVA (daily access × drug infusion × context test day), followed by post hoc comparisons. Significance was set at $p < 0.05$ for all tests and the data are presented as the mean ± SEM.

3. Results

Fig. 1 shows schematic diagrams indicating the most ventral placement of the bilateral infusion cannulae tips in the dlCPu. Animals that did not have proper bilateral placements ($n=3$) were not included in the data analysis.

Fig. 2 shows active lever responding (left) and cocaine intake (right) for animals that self-administered for 1 h/day or 6 h/day. Animals maintained at 1 h/day showed stable lever responding and cocaine intake over the entire 15 day maintenance period (i.e., no escalation over time). When analyzed across the 15 day period, animals maintained at 6 h/day showed a steady and significant increase in both daily active lever responding [$F(14,308)=1.81$, $p=0.037$] and cocaine intake [$F(14,308)=5.05$, $p<0.0001$]. Post hoc comparisons showed that both total lever responding (day 15) and cocaine intake (days 5, 6, and 8-15) were significantly increased as compared to the first day of long access. Analysis of data from the first hour of the session in the 6 h/day group showed significant increases over time in both active lever responding [$F(14,308)=1.91$, $p=0.025$] and cocaine intake [$F(14,308)=6.39$, $p<0.0001$]. Post hoc comparisons showed significantly increased cocaine intake on days 6, 8, and 10-15 when compared to day 1. Two way repeated measures ANOVA for cocaine intake during the first hour showed a significant main effect for day [$F(14,574)=4.09$, $p<0.0001$], access [$F(1,41)=12.08$, $p=0.002$], and a day $\times$ access interaction [$F(14,574)=3.03$, $p<0.0001$], with significant differences between 1 and 6 h/day access conditions at days 6, 8, and 10-15.

For context-induced cocaine-seeking, different response patterns emerged for the 1 vs. 6 h/day groups (Fig. 3). In particular, following vehicle infusions, the 1 h/day group showed the highest responding on day 1, while the 6 h/day group showed the highest responding on day 14. A three-way ANOVA showed a significant interaction of daily access $\times$ drug infusion $\times$ context test day [$F(2,100)=3.11$, $p=0.049$]. Post hoc tests showed that inactivation of the dlCPu by B/M significantly attenuated responding in the 1 h/day group on test day 1 ($p<0.05$), with no significant effects at days 14 or 60. In a similar manner, dlCPu inactivation in the 6 h/day group significantly reduced responding on day 14 ($p<0.05$), with no significant effects at days 1 or 60. In addition, for the vehicle conditions, the 1 h/day group showed higher responding on day 1 ($p<0.05$), and the 6 h/day group showed higher responding on day 14 ($p<0.05$).

4. Discussion

While both short and long access groups showed an attenuation of responding after dlCPu inactivation, animals with a greater amount of prior cocaine intake (6 h/day) did not generally show a greater dependence on intact dlCPu function during tests of cocaine-seeking. These results suggest that while the dlCPu clearly contributes to cocaine-seeking, there is not a clear pattern of an increased necessity of intact dlCPu function to maintain cocaine-seeking in animals with more prolonged histories of cocaine intake.

The long access escalation model of drug self-administration has been postulated to represent an animal model for the transition from moderate to excessive drug intake [25,27]. Growing evidence has shown that longer access to daily drug use produces enduring behavioral and neuronal changes that differ from animals with a history of short daily access. For example, drug-primed reinstatement of drug-seeking is enhanced in animals with a history of extended cocaine [18,20] or methamphetamine [28] self-administration. We saw a clear difference in cocaine intake in the two groups, with the development of escalated cocaine intake in the long, but not short access group. We also observed a different pattern of responding in the two groups when tested after abstinence, whereby the short access

group showed greater responding on the first day after self-administration and the long access group showed greater responding on day 14. However, neither group showed evidence of increased responding on day 60.

Previous reports have shown that drug-seeking induced by drug-paired cues shows an increase from immediate to later withdrawal, the so-called “incubation” of drug craving effect [22], similar to the pattern shown in our 6 h/day group. However, it is important to note that we intentionally avoided the use of an explicit discrete stimulus in the current study in order to focus on drug-seeking that emerges directly from the return to the drug context, and in keeping with our previous studies on dlCPu function after abstinence [12,13]. The absence of a greater incubation of responding, especially at 60 days of withdrawal, may be due in part to this design feature of the current study. However, it is also possible that animals with a greater number of cocaine self-administration days may have experienced a type of overtraining extinction effect, a phenomenon reported in the nondrug reinforcement literature. The overtraining extinction effect is defined as decreased resistance to extinction as a result of extended training, and may be a consequence of the experience of non-reward being more salient to animals whose training has become automatic and habitual, particularly under conditions of continuous reinforcement [29]. This effect has recently been demonstrated with cocaine self-administration under an FR1 schedule, whereby animals that received 15 days of daily 2 h sessions were more resistant to extinction than animals that received 30 days [30]. While the present study did not utilize daily extinction trials, the context tests acted as extinction sessions in that they involved unreinforced responding. Therefore, the lack of potentiated responding at abstinence day 60 in both the 1 h and 6 h access groups may have resulted from overtraining, and thus more rapid extinction of responding. Responding at abstinence day 60 following extended access cocaine potentially could be greater if animals never experienced prior context tests.

Interestingly, assessment of the role of the **dlCPu** via inactivation did not show profound differences between animals with different cocaine self-administration histories, as both groups depended on the intact function of the dlCPu on days 1 and 14 of abstinence, with the greater inactivation effect apparent on day 1 in the 1 h access group (69% decrease from vehicle) and on day 14 in the 6 h access group (78% decrease from vehicle). While the current results do not support the contention that greater intake of cocaine leads to a greater dependence on dlCPu function, our study has some inherent limitations. Specifically, we intentionally avoided the use of explicit cocaine-paired cues during both the self-administration phase and the subsequent context tests in order to avoid the contribution of explicit, contingent cue presentations, which are known to come under the control of amygdalar and prefrontal cortical pathways [31]. It may be the case that prolonged cocaine self-administration alters dorsal striatal mediation of discrete cue-maintained cocaine-seeking [11]. However, it should be noted that the dlCPu has been shown to play a clear role in context-induced cocaine [12] and heroin [32] seeking. Secondly, in order to maximize the amount of behavioral testing obtained in each rat, we tested animals on three separate context tests. We did not see any evidence for order effects and animals in both access groups underwent the same procedures. However, repeated testing at the three time points may have affected our results by a general reduction in responding over time, particularly at day 60. Finally, our design did not incorporate daily extinction trials between the context tests. Since extensive extinction training can produce neuroadaptations that differ from those seen after abstinence [33], the role of the dlCPu at different withdrawal timepoints may differ in an extinction-reinstatement model.

Understanding the neural pathways of cocaine addiction, specifically those that underlie cocaine-seeking at the time of relapse, is critical to the ultimate development of successful addiction treatments. Although converging evidence points to a previously under recognized

role of dorsal striatal pathways in the mediation of cocaine-seeking after prolonged cocaine abuse, the contribution of the dorsal striatum appears to be more complex than a simple cumulative intake dependent process.

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References

1. Taylor JR, Robbins TW. 6-Hydroxydopamine lesions of the nucleus accumbens, but not of the caudate nucleus, attenuate enhanced responding with reward-related stimuli produced by intra-accumbens d-amphetamine. *Psychopharmacology* 1986;90:390–7. [PubMed: 3097729]
2. Kuhar MJ, Ritz MC, Boja JW. The dopamine hypothesis of the reinforcing properties of cocaine. *Trends Neurosci* 1991;14:299–302. [PubMed: 1719677]
3. Koob GF. Drugs of abuse: anatomy, pharmacology and function of reward pathways. *Trends Pharmacol Sci* 1992;13:177–84. [PubMed: 1604710]
4. Canales JJ. Intermittent cortical stimulation evokes sensitization to cocaine and enduring changes in matrix and striosome neuron responsiveness. *Synapse* 2005;57:56–60. [PubMed: 15858832]
5. Everitt BJ, Robbins TW. Neural systems of reinforcement for drug addiction: from actions to habits to compulsion. *Nat Neurosci* 2005;8:1481–9. [PubMed: 16251991]
6. Gerdeman GL, Partridge JG, Lupica CR, Lovinger DM. It could be habit forming: drugs of abuse and striatal synaptic plasticity. *Trends Neurosci* 2003;26:184–92. [PubMed: 12689769]
7. Porrino LJ, Lyons D, Smith HR, Daunais JB, Nader MA. Cocaine self-administration produces a progressive involvement of limbic, association, and sensorimotor striatal domains. *J Neurosci* 2004;24:3554–62. [PubMed: 15071103]
8. Belin D, Jonkman S, Dickinson A, Robbins TW, Everitt BJ. Parallel and interactive learning processes within the basal ganglia: relevance for the understanding of addiction. *Behav Brain Res* 2009;199:89–102. [PubMed: 18950658]
9. Ito R, Dalley JW, Robbins TW, Everitt BJ. Dopamine release in the dorsal striatum during cocaine-seeking behavior under the control of a drug-associated cue. *J Neurosci* 2002;22:6247–53. [PubMed: 12122083]
10. Vanderschuren LJ, Di Ciano P, Everitt BJ. Involvement of the dorsal striatum in cue-controlled cocaine seeking. *J Neurosci* 2005;25:8665–70. [PubMed: 16177034]
11. Belin D, Everitt BJ. Cocaine seeking habits depend upon dopamine-dependent serial connectivity linking the ventral with the dorsal striatum. *Neuron* 2008;57:432–41. [PubMed: 18255035]
12. Fuchs RA, Branham RK, See RE. Different neural substrates mediate cocaine seeking after abstinence versus extinction training: a critical role for the dorsolateral caudate-putamen. *J Neurosci* 2006;26:3584–8. [PubMed: 16571766]
13. See RE, Elliott JC, Feltenstein MW. The role of dorsal vs ventral striatal pathways in cocaine-seeking behavior after prolonged abstinence in rats. *Psychopharmacology* 2007;194:321–31. [PubMed: 17589830]
14. Letchworth SR, Nader MA, Smith HR, Friedman DP, Porrino LJ. Progression of changes in dopamine transporter binding site density as a result of cocaine self-administration in rhesus monkeys. *J Neurosci* 2001;21:2799–807. [PubMed: 11306632]
15. Volkow ND, Wang GJ, Telang F, Fowler JS, Logan J, Childress AR, Jayne M, Ma Y, Wong C. Cocaine cues and dopamine in dorsal striatum: mechanism of craving in cocaine addiction. *J Neurosci* 2006;26:6583–8. [PubMed: 16775146]
16. Wong DF, Kuwabara H, Schretlen DJ, Bonson KR, Zhou Y, Nandi A, Brasic JR, Kimes AS, Maris MA, Kumar A, Contoreggi C, Links J, Ernst M, Rousset O, Zukin S, Grace AA, Lee JS, Rohde C, Jasinski DR, Gjedde A, London ED. Increased occupancy of dopamine receptors in human

- striatum during cue-elicited cocaine craving. *Neuropsychopharmacology* 2006;31:2716–27. [PubMed: 16971900]
17. Crombag HS, Grimm JW, Shaham Y. Effect of dopamine receptor antagonists on renewal of cocaine seeking by reexposure to drug-associated contextual cues. *Neuropsychopharmacology* 2002;27:1006–15. [PubMed: 12464457]
 18. Kippin TE, Fuchs RA, See RE. Contributions of prolonged contingent and noncontingent cocaine exposure to enhanced reinstatement of cocaine seeking in rats. *Psychopharmacology* 2006;187:60–7. [PubMed: 16598453]
 19. Knackstedt LA, Kalivas PW. Extended access to cocaine self-administration enhances drug-primed reinstatement but not behavioral sensitization. *J Pharmacol Exp Ther* 2007;322:1103–9. [PubMed: 17601982]
 20. Mantsch JR, Yuferov V, Mathieu-Kia AM, Ho A, Kreek MJ. Effects of extended access to high versus low cocaine doses on self-administration, cocaine-induced reinstatement and brain mRNA levels in rats. *Psychopharmacology* 2004;175:26–36. [PubMed: 15042275]
 21. Neisewander JL, Baker DA, Fuchs RA, Tran-Nguyen LT, Palmer A, Marshall JF. Fos protein expression and cocaine-seeking behavior in rats after exposure to a cocaine self-administration environment. *J Neurosci* 2000;20:798–805. [PubMed: 10632609]
 22. Grimm JW, Hope BT, Wise RA, Shaham Y. Neuroadaptation. Incubation of cocaine craving after withdrawal. *Nature* 2001;412:141–2. [PubMed: 11449260]
 23. Freeman WM, Patel KM, Brucklacher RM, Lull ME, Erwin M, Morgan D, Roberts DC, Vrana KE. Persistent alterations in mesolimbic gene expression with abstinence from cocaine self-administration. *Neuropsychopharmacology* 2008;33:1807–17. [PubMed: 17851536]
 24. Paxinos, G.; Watson, C. The rat brain in stereotaxic coordinates. New York: Academic Press; 1997.
 25. Ahmed SH, Koob GF. Transition from moderate to excessive drug intake: change in hedonic set point. *Science* 1998;282:298–300. [PubMed: 9765157]
 26. Ahmed SH, Koob GF. Changes in response to a dopamine receptor antagonist in rats with escalating cocaine intake. *Psychopharmacology* 2004;172:450–4. [PubMed: 14647962]
 27. Ahmed SH, Kenny PJ, Koob GF, Markou A. Neurobiological evidence for hedonic allostasis associated with escalating cocaine use. *Nat Neurosci* 2002;5:625–6. [PubMed: 12055635]
 28. Rogers JL, De Santis S, See RE. Extended methamphetamine self-administration enhances reinstatement of drug seeking and impairs novel object recognition in rats. *Psychopharmacology* 2008;199:615–24. [PubMed: 18493748]
 29. Tombaugh TN. The overtraining extinction effect with a discrete-trial bar-press procedure. *J Exp Psychol* 1967;73:632–4. [PubMed: 6034023]
 30. Valles R, Rocha A, Nation JR. The effects of acquisition training schedule on extinction and reinstatement of cocaine self-administration in male rats. *Exp Clin Psychopharmacol* 2006;14:245–53. [PubMed: 16756428]
 31. Feltenstein MW, See RE. The neurocircuitry of addiction: an overview. *Br J Pharmacol* 2008;154:261–74. [PubMed: 18311189]
 32. Bossert JM, Wihbey KA, Pickens CL, Nair SG, Shaham Y. Role of dopamine D(1)-family receptors in dorsolateral striatum in context-induced reinstatement of heroin seeking in rats. *Psychopharmacology* 2009;206:51–60. [PubMed: 19506837]
 33. Knackstedt LA, Moussawi K, Lalumiere R, Schwendt M, Klugmann M, Kalivas PW. Extinction training after cocaine self-administration induces glutamatergic plasticity to inhibit cocaine seeking. *J Neurosci* 2010;30:7984–92. [PubMed: 20534846]

Fig. 1. Schematic illustration of cannula tip placements in the dlCPu (adapted from Paxinos and Watson, 1997). Numbers to the right of the coronal sections indicate anterior distance (mm) from bregma.

Fig. 2.

Cocaine self-administration (1 h/day and 6 h/day) as seen for: A) total active lever responses, B) total cocaine intake (mg/kg), C) active lever responses in the first hour, D) cocaine intake in the first hour. Data are expressed as mean $\pm$ SEM. Significant differences relative to day 1 are indicated (* p <0.05; ** p <0.01).

Fig. 3.

Inactivation of the dlCPu and cocaine-seeking after various periods of abstinence.

Responses (mean±SEM) on the previously cocaine-paired lever are shown after bilateral infusion of vehicle or B/M just before context test days 1, 14, and 60 in the 1 h/day group (n=7-10 per condition) and in the 6 h/day group (n=8-12 per condition). Significant differences are indicated between vehicle and B/M infusion (* p <0.05) and between 1 h and 6 h/day groups († p <0.05) for context test days 1 and 14.